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Sulcispora supratumida sp. nov. (Phaeosphaeriaceae, Pleosporales) on *Anthoxanthum odoratum* from Italy

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Abstract

Sulcispora is typified by *S. pleurospora*. We collected a sulcispora-like taxon on leaves of *Anthoxanthum odoratum* L. in Italy and obtained single ascospore isolates. Combined ITS, LSU, SSU and tef1 sequence analyses suggested that *Sulcispora* is placed in the family Phaeosphaeriaceae and a newly collected *Sulcispora* species is introduced here as *S. supratumida* sp. nov. Detailed descriptions and illustrations are provided for *Sulcispora supratumida* and it is compared with the type species, *S. pleurospora*.

Keywords

Combined gene analysis, Dothideomycetes, graminicolous fungi, new species, spore septation

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Introduction

Phaeosphaeriaceae is a highly diverse and large family in the order Pleosporales (Hyde et al. 2013) with more than 42 accepted genera (Hyde et al. 2017; Karunarathna et al. 2017; Wanasinghe et al. 2018). Members of Phaeosphaeriaceae are pathogens or hyper-parasites on living plants and humans and saprobes of decaying plant matter (Tennakoon et al. 2016; Ahmed et al. 2017).

Sulcispora was proposed by Shoemaker and Babcock (1989) as a monotypic genus to accommodate *Sulcispora pleurospora* (≡ *Phaeosphaeria pleurospora* Niessl). Some morphological characters of *Phaeosphaeria pleurospora* did not fit within species concepts of *Phaeosphaeria* and Shoemaker and Babcock (1989), therefore, introduced the genus *Sulcispora*. The genus name refers to the numerous furrows on the ascospore wall (Shoemaker and Babcock 1989). *Sulcispora pleurospora* has been reported on monocotyledonous hosts in genera such as *Anthoxanthum*, *Carex*, *Deschampsia*, *Sesleria* and *Tofieldia* (Leuchtmann 1984; Shoemaker and Babcock 1989).

In this study, we collected sulcispora-like species associated with leaf spots of *Anthoxanthum odoratum* in Italy. We compared the morphological characters of our collection with the isotype of *Sulcispora pleurospora*. Morphologically, our collection differs from the type species of *Sulcispora*, *S. pleurospora*. Therefore, we introduce our collection as a new species. Combined ITS, LSU, SSU and tef1 sequence analysis including taxa in Phaeosphaeriaceae indicates that the here-studied fungus grouped with "*Phaeosphaeria pleurospora*" (CBS 460.84) with high support value.

Methods

Sample collection, specimen examination and single spore isolation

Specimens were collected from *Anthoxanthum odoratum* L. from Italy in 2013. They were examined and photographed using a Carl Zeiss Discovery V8 stereo-microscope fitted with Axiocam. Sections of ascomata were taken by hand under a stereo-microscope. Sections and other micro-morphological characters were photographed using a Nikon Eclipse 80i compound microscope fitted with a Canon 450D digital camera. All microscopic measurements were made with Tarosoft image framework (v. 0.9.0.7). Colony characteristics were recorded from cultures grown on Malt Extract Agar (MEA).

Single spore isolation was carried out following the method described by Chomnunti et al. (2014). Germinated ascospores were aseptically transferred into fresh MEA plates and incubated at 20 °C to obtain pure cultures and later transferred to MEA slants and stored at 4 °C for further study. The holotype and paratype specimens were deposited at the Mae Fah Luang University (MFLU) fungaria and the herbarium of Kunming Institute of Botany, Chinese Academy of Sciences (HKAS), respectively. Living cultures were deposited at the Mae Fah Luang Culture Collection (MFLUCC).

MycoBank (http://www.mycobank.org/) and Facesoffungi (Jayasiri et al. 2015) numbers were obtained for the new strain. The new species was established based on recommendations outlined by Jeewon and Hyde (2016).

DNA extraction, PCR amplification and DNA sequencing

Fresh fungal mycelium grown on MEA for four weeks at 20°C was used for DNA extraction (Jeewon et al. 2002). Genomic DNA extraction and PCR reactions were carried out using ITS4/ITS5 for internal transcribed spacer nrDNA (ITS), LR5/LROR for large subunit nrDNA (LSU), NS1/NS4 for large subunit nrDNA (SSU) and 983F/2218R for translation elongation factor 1 (tef1) genes according to the same protocol of Maharachchikumbura et al. (2012). The PCR products were observed on 1% agarose electrophoresis gel stained with ethidium bromide. Purification and sequencing of PCR products were carried out at the Kunming Institute of Botany, Chinese Academy of Science, Kunming, China. Sequence quality was checked and sequences were condensed with DNASTAR Lasergene v.7.1. Sequences derived in this study were deposited in GenBank (Table 1).

Sequence alignment and phylogenetic analysis

BLASTn searches were made using the newly generated sequences to assist in taxon sampling for phylogenetic analyses. In addition, representatives of the Phaeosphaeriaceae were selected following Tennakoon et al. (2016) and Wanasinghe et al. (2018) (Table 1). Combined multi-locus sequence data of ITS, LSU, SSU and tef1 regions were aligned using default settings of MAFFT v.7 (Katoh et al. 2017) and manually adjusted using BioEdit 7.1.3 (Hall 1999) to allow maximum alignment and minimum gaps. Maximum likelihood analysis was performed by RAxML (Stamatakis and Alachiotis 2010) implemented in raxmlGUIv.1.3 (Silvestro and Michalak 2012). The search strategy was set to rapid bootstrapping and the analysis carried out using the GTRGAMMAI model of nucleotide substitution with 1000 replicates. The model of evolution was estimated by using MrModeltest 2.2 (Nylander 2004).

For the Bayesian inference (BI) analyses of the individual loci and concatenated ITS, LSU, SSU and tef-1 alignment, the above mentioned model test was used to determine the best fitting nucleotide substitution model settings for MrBayes v. 3.0b4. A dirichlet state frequency was predicted for all three data partitions and GTR+I+G as the best model for all single gene and combined datasets. The heating parameter was set to 0.2 and trees were saved every 1000 generations (Ronquist and Huelsenbeck 2003). The Markov Chain Monte Carlo (MCMC) analysis of four chains started in parallel from a random tree topology. The Bayesian analysis lasted 10,000,000 generations (average standard deviation of split frequencies value = 0.0098) and the consensus trees and posterior probabilities were calculated from the 9,998,000 trees sampled

Table 1. Isolates used in this study and their GenBank and culture accession numbers. The strain of *Sulcispora supratumida* sp. nov. is set in bold font and all ex-type strains are annotated with "T".

Taxon	Culture accession no	ITS	LSU	SSU	tef-1
Allophaeosphaeria muriformia	MFLUCC 13-0349^T	KP765680	KP765681	KP765682	–
A. subcylindrospora	MFLUCC 13-0380^T	KT314184	KT314183	KT314185	–
Amarenographium ammophilae	MFLUCC 16-0296^T	KU848196	KU848197	KU848198	MG520894
Ampelomyces quisqualis	CBS 129.79^T	HQ108038	JX681064	EU754029	–
Bhatiellae rosae	MFLUCC 17-0664^T	MG828873	MG828989	MG829101	–
Chaetosphaeronema hispidulum	CBS 216.75	KF251148	KF251652	EU754045	–
Dactylidina dactylidis	MFLUCC 14-0963^T	MG828887	MG829003	MG829114	MG829199
D. shoemakeri	MFLUCC 14-0966^T	MG828886	MG829002	MG829113	MG829200
Dematiopleospora mariae	MFLUCC 13-0612^T	–	KJ749653	KJ749652	KJ749655
Didymella exigua	CBS 183.55^T	GU237794	EU754155	EU754056	–
Didymocyrtis caloplacae	CBS 129338	JQ238641	JQ238643	–	–
D. ficuzzae	CBS 128019	KP170647	JQ238616	–	–
D. cladoniicola	CBS 128026	JQ238626	–	–	–
Embarria clematidis	MFLUCC 14-0976^T	MG828871	MG828987	MG829099	MG829194
Entodesmium rude	CBS 650.86	–	GU301812	–	GU349012
Equiseticola fusispora	MFLUCC 14-0522^T	KU987668	KU987669	KU987670	MG520895
Galliicola pseudophaeosphaeria	MFLUCC 14-0527^T	KT326692	KT326693	–	MG829203
Hawksworthiana clematidicola	MFLUCC 14-0910^T	MG828901	MG829011	MG829120	MG829202
H. lonicerae	MFLUCC 14-0955^T	MG828902	MG829012	MG829121	MG829203
Italica achilleae	MFLUCC 14-0959^T	MG828903	MG829013	MG829122	MG829204
Juncaceicola alpine	CBS 456.84	KF251181	KF251684	–	–
J. luzulae	MFLUCC 16-0780	KX449529	KX449530	KX449531	MG520898
Leptospora rubella	CPC 11006	DQ195780	DQ195792	DQ195803	–
Loratospora aestuarii	JK 5535B	–	GU301838	GU296168	–
L. luzulae	MFLUCC 14-0826	KT328497	KT328495	KT328496	–
Melnikia anthoxanthii	MFLUCC 14-1010^T	KU848205	KU848204	–	–
Muriphaeosphaeria galatellae	MFLUCC 14-0614^T	KT438333	KT438329	KT438331	MG520900
Neosetophoma italica	MFLUCC14-0826^T	KP711356	KP711361	KP711366	–
N. samarorum	CBS 138.96^T	FJ427061	KF251664	GQ387517	–
Neostagonospora caricis	CBS 135092/S616^T	KF251163	KF251667	–	–
N. eligiae	CBS 135101^T	KF251164	KF251668	–	–
Nodulosphaeria hirta	MFLUCC 13-0867	KU708849	KU708845	KU708841	KU708853
N. senecionis	MFLUCC 15-1297	KT290257	KT290258	KT290259	–
Ophiobolus cirsii	MFLUCC 13-0218^T	KM014664	KM014662	KM014663	–
O. disseminans	AS2L14-6	–	–	KP117305	–
Ophiosphaerella agrostidis	MFLUCC 11-0152^T	KM434271	KM434281	KM434290	KM434299
Paraleptosphaeria dryadis	CBS 643.86	J F740213	GU301828	KC584632	GU349009
Paraphoma chrysanthemicola	CBS 522.66	FJ426985	KF251670	GQ387521	–
P. radicina	CBS 111.79^T	KF251172	KF251676	EU754092	–
Parastagonospora nodorum	CBS 110109^T	KF251177	KF251681	EU754076	–
P. poagena	CBS 136776^T	KJ869116	KJ869174	–	–
Phaeosphaeria chiangraina	MFLUCC 13-0231^T	KM434270	KM434280	KM434289	KM434298
P. oryzae	CBS 110110^T	KF251186	KF251689	GQ387530	–
P. papayae	S528	KF251187	KF251690	–	–

Taxon	Culture accession no	ITS	LSU	SSU	tef-1
Phaeosphaeria pleurospora	CBS 460.84	AF439498	–	–	–
Phaeosphaeriopsis glaucopunnctata	MFLUCC 13-0265^T	KJ522473	KJ522477	KJ522481	MG520918
P. triseptata	MFLUCC 13-0271^T	KJ522475	KJ522479	KJ522484	MG520919
Poaceicola arundinis	MFLUCC 15-0702^T	KU058716	KU058726	–	MG520921
P. italica	MFLUCC 13-0267^T	KX926421	KX910094	KX950409	MG520924
Populocrescntia forlicesesensis	MFLU 15-0651^T	KT306948	KT306952	KT306955	MG520925
Premilcurensis senecionis	MFLUCC 13-0575^T	KT728365	KT728366	–	–
Sclerostagonospora sp.	CBS 123538	FJ372393	FJ372410	–	–
Scolicosporium minkeviciusii	MFLUCC 12-0089^T	–	KF366382	KF366383	–
Septoriella leuchtmannii	CBS 459.84^T	KF251188	KF251691	–	–
Setomelanomma holmii	CBS 110217	–	GU301871	GQ387572	GU349028
Setophoma sacchari	CBS 333.39^T	KF251245	KF251748	GQ387525	–
S. terrestris	CBS 335.29^T	KF251246	KF251749	GQ387526	–
Sulcispora supratumida	**MFLUCC 14-0995**	**KP271443**	**KP271444**	**KP271445**	**MH665366**
Tintelnotia destructans	CBS 127737^T	NR_147684	NG_058274	KY090698	–
T. destructans	CBS 137534	–	KY090663	KY090697	–
Vagicola chlamydospora	MFLUCC 15-0177^T	KU163658	KU163654	–	–
V. vagans	CBS 604.86	KF251193	KF251696	–	–
Vrystaatia aloeicola	CBS 135107	KF251278	KF251781	–	–
Wojnowicia dactylidis	MFLUCC 13-0735^T	KP744470	KP684149	KP684150	–
W. lonicerae	MFLUCC 13-0737^T	KP744471	KP684151	KP684152	–
Wojnowiciella eucalypti	CPC 25024^T	KR476741	KR476774	–	LT990617
Xenoseptoria neosaccardoi	CBS 128665^T	KF251281	KF251784	–	–
X. neosaccardoi	CBS 120.43	KF251280	KF251783	–	–
Yunnanensis phragmitis	MFLUCC 17-0315^T	MF684862	MF684863	MF684867	MF683624
Y. phragmitis	MFLUCC 17-1365^T	MF684869	MF684865	MF684864	MF683625

CBS: Westerdijk Fungal Biodiversity Institute, Utrecht, The Netherlands; **CPC**: Culture collection of Pedro Crous, housed at CBS-KNAW; **MFLUCC**: Mae Fah Luang University Culture Collection, Chiang Rai, Thailand.

after discarding the first 20% of generations as burn-in. Trees obtained in this study were deposited in TreeBASE under accession number S22938. The phylogram was visualised in FigTree v. 1.2.2 (Rambaut and Drummond 2008).

Results

Phylogenetic inferences

The combined ITS, LSU, SSU and tef-1 sequence data set comprised 69 strains of Phaeosphaeriaceae with *Didymella exigua* as the outgroup taxon. All individual trees generated under different criteria and from single gene datasets were essentially similar in topology and not significantly different from the tree generated from the concat-

enated dataset. Maximum likelihood analysis with 1000 bootstrap replicates yielded a tree with the likelihood value of ln: -13019.593920 and the following model parameters: alpha: 0.144187; ∏(A): 0.245356, ∏(C): 0.229408, ∏(G): 0.267562 and ∏(T): 0.257674. The best scoring RAxML tree is shown in Figure 1. Maximum likelihood bootstrap values ≥50% and Bayesian inference (BI) ≥0.9 are given at each node.

The phylogenetic trees obtained from maximum likelihood were topologically congruent to previous studies on Phaeosphaeriaceae (Phookamsak et al. 2014; Thambugala et al. 2014; Tennakoon et al. 2016; Karunarathna et al. 2017; Wanasinghe et al. 2018). This phylogenetic analysis showed the placement of 45 genera within Phaeosphaeriaceae. The here-studied strain clustered with CBS 460.84 (one of Leuchtmann's Swiss strains of *S. pleurospora* from *Carex firma*) with 100% bootstrap support value. The ITS sequence of the CBS 460.84 is almost identical to our strain (MFLUCC 14–0995). However no LSU, SSU and tef-1 sequences were obtained from CBS 460.84 in GenBank. The herbarium specimen of CBS 460.84 is in Westerdijk Fungal Biodiversity Institute (CBS) under accession number CBS H-15991 (SWITZERLAND, Kt. Graubünden, Zügenschlucht near Davos, *Carex firma*, A. Leuchtmann). However, CBS has presently stopped sending specimens on loan, hence we could not compare morphological characters of the here studied strain with CBS 460.84. Additionally *Sulcispora* sisterly clustered with the type species of *Loratospora*, *L. aestuarii* with low support and the second species of *Loratospora*, *L. luzulae*. was distantly clustered.

Taxonomy

Sulcispora supratumida **Senan., Camporesi & K.D. Hyde, sp. nov.**
MycoBank No: MB826887
Facesoffungi No: FoF 04782
Figure 2

Etymology. The species epithet is based on the two Latin words "supra" meaning upper and "tumidus" meaning swollen, referring to the position of swollen cells of ascospores.

Type. ITALY. Province of Forli-Cesena, Premilcuore, Passodella Valbura, on dead leaves of *Anthoxanthum odoratum* L. (Poaceae), 25 May 2013, Erio Camporesi, IT 1306 (MFLU 15–0038, holotype; HKAS 83865, paratype): living cultures, MFLUCC 14–0995.

Description. *Saprobic* on leaves of *Anthoxanthum odoratum* L., visible as black spots, occurring on the upper surface of entire leaf. *Sexual morph*. *Ascomata* 110–150 × 90–140 µm ($\bar{x}$ = 140–125 µm, n = 10), scattered, solitary, immersed, uniloculate, globose, black. *Ostiole* 35–40 µm ($\bar{x}$ = 39 µm, n = 10) wide, papillate, central, periphysate. *Periphyses* 15–20 µm long, hyaline. *Peridium* comprising 2–4 layers of brown to dark brown, thick-walled, cells of *textura angularis* to *textura globularis*. *Hamathecium* comprising

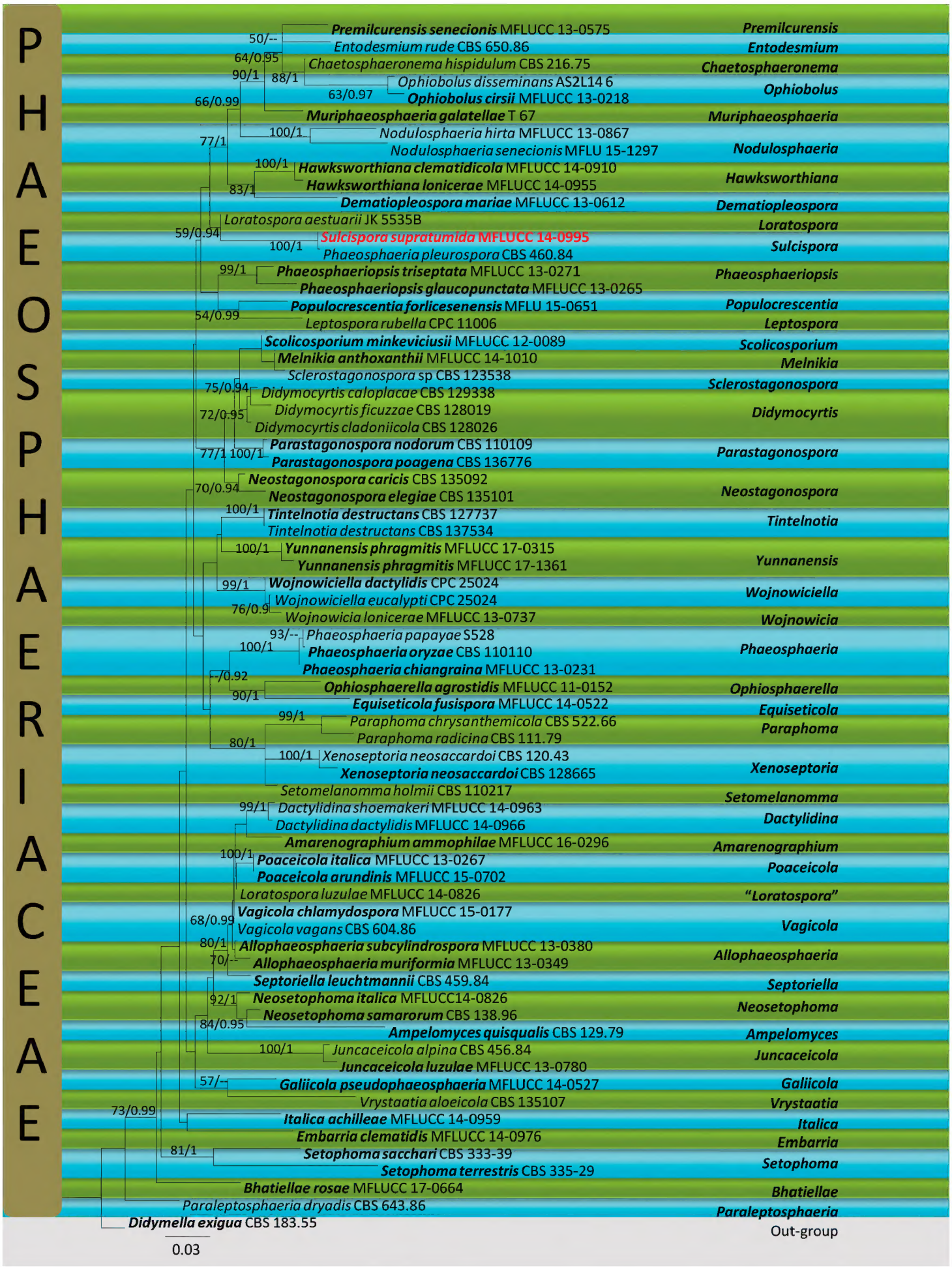

Figure 1. Maximum likelihood majority rule consensus tree based on a combined dataset of ITS, LSU, SSU and tef-1 sequences. Bootstrap support values ≥50% and Bayesian inference (BI) ≥0.9 are given at the nodes. The tree is rooted to *Didymella exigua* (CBS 183.55). The culture accession numbers are given after the species names. All ex-type strains are in bold. The newly introduced species from this study is in bold red.

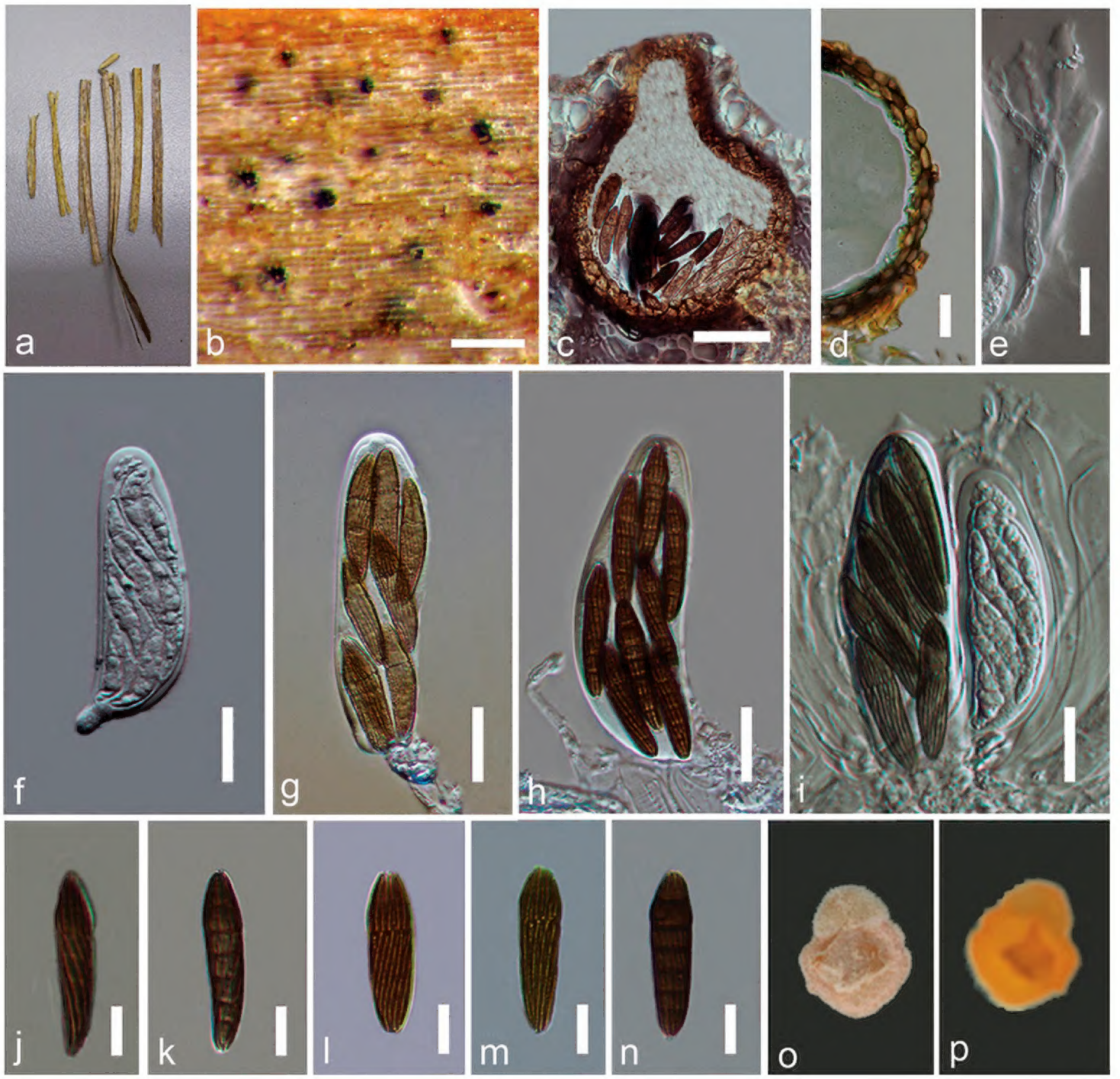

Figure 2. *Sulcispora supratumida* (MFLU 15–0038). **a** Leaves of *Anthoxanthum odoratum* **b** Appearance of ascomata on host surface **c** Cross section of ascoma **d** Peridium **e** Pseudoparaphyses **f–i** Asci **j–n** Ascospores **o** Upper surface of the culture **p** Lower surface of the culture. Scale bars: 200 μm (**b**), 50 μm (**c**), 20 μm (**d–i**), 10 μm (**j–n**).

2–4 μm wide, cellular, hyaline, branched, septate, pseudoparaphyses, constricted at the septa, anastomosing mostly above the asci and embedded in a mucilaginous matrix. *Asci* 85–125 × 20–35 μm ($\overline{x}$ = 100 × 30 μm, n = 20), 8-spored, few, bitunicate, fissitunicate, subglobose to clavate, short pedicellate, apically rounded, with an ocular chamber, arising from the base of the ascoma and attached to parenchymatous cell matrix at base. *Ascospores* 30–35 × 6–9 μm ($\overline{x}$ = 35 × 7 μm, n = 25), bi-seriate to tri-seriate, narrowly fusiform, narrowing towards the end cells, reddish to dark brown, 6-septate, second septum supra-median, slightly constricted, not constricted at other septa, second segment swollen, straight, with 12–16 longitudinal furrows on surface, lacking a mucilaginous sheath. *Asexual morph*. Undetermined.

Table 2. Ascospore morphology comparison of *Sulcispora* species

Species name	Herbarium type data	Host	No of septa	Swollen cell	Reference
Sulcispora pleurospora	FH 196419 (isotype)	*Deschampsia cespitosa* (Poaceae)	5–6	3rd	Shoemaker and Babcock 1989
	F6952, F6949, F6951 (isotype)	*Deschampsia cespitosa* (Poaceae)	6	3rd	In this study
	M (1 collection), ZT (8 collections)	6 monocotyledonous hosts, 1 dicotyledonous host	6–8	3rd or 4th	Leuchtmann 1984
Sulcispora supratumida	ZT (6 collections)	*Seleria caerulea* (Poaceae) *Carex firma* (Cyperaceae)	6	2nd	Leuchtmann 1984
	MFLU 15-0038 (holotype)	*Anthoxanthum odoratum* (Poaceae)	6	2nd	In this study

Culture characteristics. 2 cm diameter after 4 weeks incubated in dark at 25 °C on MEA, pinkish-white, circular, slightly woolly, margin lobate, effuse, lacking aerial mycelium, tightly attached to the media.

Discussion

Shoemaker and Babcock (1989) observed type specimens of *Phaeosphaeria pleurospora* and found that the ascospores of *P. pleurospora* with striated ornamented walls are different to those of other genera in Phaeosphaeriaceae. Hence, they introduced the genus *Sulcispora* to accommodate *P. pleurospora* and placed it in Phaeosphaeriaceae. *Sulcispora pleurospora* has some similarities with *Phaeosphaeria exarata* Shoemaker & C.E. Babc., in having very large cells in the peridium, ascospores with a continuous sheath and ornamented wall of ascospores with coarse, longitudinal ridges (Shoemaker and Babcock 1989).

In this study, a combined gene sequence analysis of taxa amongst the Phaeosphaeriaceae provides substantial evidence to support *Sulcispora* as a distinct genus in Phaeosphaeriaceae. *Sulcispora* differs from other genera in having immersed ascomata with a relatively thin wall, cellular pseudoparaphyses, short pedicellate asci and brown ascospores (Phookamsak et al. 2014).

Leuchtmann (1984) reported variation of ascospore septation amongst several collections of *Phaeosphaeria pleurospora* from different host plants. *Phaeosphaeria pleurospora,* collected from *Sesleria caerulea* (L.) Ard. and *Carex firma* Mygind ex Host, usually formed 6-septate ascospores and the second segment was swollen. Our collection is morphologically identical to Leuchtmann's collection. However, the isotype and some of Leuchtmann's collections from other host plants had 5–8-septate ascospores and the third or fourth segment was swollen (Table 2). Therefore Leuchtmann (1984) characterised *Phaeosphaeria pleurospora* as a species with 5–8 septate ascospores. However, Leuchtmann's collection of *Sulcispora pleurospora* is likely to comprise more than a single species and possibly constitutes a species complex.

Based on the morphology, we identified our collection as different from the isotype of *Sulcispora pleurospora*. Hence, we introduced a new species as *Sulcispora supratumida* sp. nov. However, the ITS sequence of our strain clustered with that of CBS 460.84 (one of Leuchtmann's Swiss strain of *S. pleurospora* from *Carex firma*) with 100% bootstrap support value. There are only two base pair differences between the ITS regions of both strains. Since there are no sequence data of other DNA regions of *Sulcispora pleurospora* deposited in GenBank, we could not confirm whether or not CBS 460.84 is *Sulcispora supratumida*. However, it would eventually be practical to obtain the living strain of CBS 460.84 and generate further sequence data.

Keys for species in *Sulcispora*

1 Ascomata erumpent, long papillate, 5–8-septated, ascospores with 3rd swollen cell ..... ***S. pleurospora***

– Ascomata immersed, short papillate, 6-septated, ascospores with 2nd swollen cell ..... ***S. supratumida***

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References

Ahmed SA, Hofmüller W, Seibold M, de Hoog GS, Harak H, Tammer I, van Diepeningen AD, Behrens-Baumann W (2017) *Tintelnotia*, a new genus in Phaeosphaeriaceae harbouring agents of cornea and nail infections in humans. Mycoses 60(4): 244–253. http://dx.doi.org/10.1111/myc.12588

Chomnunti P, Hongsanan S, Aguirre-Hudson B, Tian Q, Peršoh D, Dhami MK, Alias AS, Xu J, Liu X, Stadler M, Hyde KD (2014) The sooty moulds. Fungal Diversity 66: 1–36. https://doi.org/10.1007/s13225-014-0278-5

Hall TA (1999) BioEdit: a user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT. Nucleic Acids Symposium Series 41: 95–98. http://brownlab.mbio.ncsu.edu/JWB/papers/1999Hall1.pdf

Hyde KD, Jones EBG, Liu JK, Ariyawansa HA, Boehm E, Boonmee S, Braun U, Chomnunti P, Crous PW, Dai DQ, Diederich P, Dissanayake A, Doilom M, Doveri F, Hongsanan S, Jayawardena R, Lawrey JD, Li YM, Liu YX, Lücking R, Monkai J, Muggia L, Nelsen MP, Pang KL, Phookamsak R, Senanayake IC, Shearer CA, Suetrong S, Tanaka K, Thambugala KM, Wijayawardene NN, Wikee S, Wu HX, Zhang Y, Aguirre-Hudson B, Alias SA, Aptroot A, Bahkali A, Bezerra JL, Bhat DJ, Camporesi E, Chukeatirote E, Gueidan C,

Hawksworth DL, Hirayama K, Hoog SD, Kang JC, Knudsen K, Li WJ, Li XH, Liu ZY, Mapook A, Mckenzie EHC, Miller AN, Mortimer PE, Phillips AJL, Raja HA, Scheuer C, Schumm F, Taylor JE, Tian Q, Tibpromma S, Wanasinghe DN, Wang Y, Xu JC, Yacharoen S, Yan JY, Zhang M (2013) Families of Dothideomycetes. Fungal Diversity 63: 1–313. http://dx.doi.org/10.1007/s13225-013-0263-4

Hyde KD, Norphanphoun C, Abreu VP, Bazzicalupo1 A, Chethana KWT, Clericuzio M, Dayarathne MC, Dissanayake AJ, Ekanayaka1 AH, He MQ, Hongsanan S, Huang SK, Jayasiri SC, Jayawardena RS, Karunarathna A, Konta S, Kusan I, Lee H, Li J, Lin CG, Liu NG, Lu YZ, Luo ZL, Manawasinghe IS, Mapook A, Perera RH, Phookamsak R, Phukhamsakda C, Siedlecki I, Soares AM, Tennakoon DS, Tian Q, Tibpromma S, Wanasinghe DN, Xiao YP, Yang J, Zeng XY, Abdel-Aziz FA, Li WJ, Senanayake IC, Shang QJ, Daranagama DA, de Silva NI, Thambugala KM, Abdel-Wahab MA, Bahkali AH, Berbee ML, Boonmee S, Bhat DJ, Bulgakov TS, Buyck B, Camporesi E, Castaneda-Ruiz RF, Chomnunti P, Doilom M, Dovana F, Gibertoni TB, Jadan M, Jeewon R, Jones EBG, Kang JC, Karunarathna SC, Lim YW, Liu JK, Liu ZY, Plautz Jr. HL, Lumyong S, Maharachchikumbura SSN, Matocec N, McKenzie EHC, Mesic A, Miller D, Pawłowska J, Pereira OL, Promputtha I, Romero AL, Ryvarden L, Su HY, Suetrong S, Tkalcec Z, Vizzini A, Wen TC, Wisitrassameewong K, Wrzosek M, Xu JC, Zhao Q, Zhao RL, Mortimer PE (2017) Fungal diversity notes 603–708: taxonomic and phylogenetic notes on genera and species. Fungal Diversity 87: 1–235. https://doi.org/10.1007/s13225-017-0391-3

Jayasiri SC, Hyde KD, Ariyawansa HA, Bhat J, Buyck B, Cai L, Dai YC, Abd-Elsalam KA, Ertz D, Hidayat I, Jeewon R, Jones EBG, Bahkali AH, Karunarathna SC, Liu JK, Luangsa-ard JJ, Lumbsch HT, Maharachchikumbura SSN, McKenzie EHC, Moncalvo JM, Ghobad-Nejhad M, Nilsson H, Pang KA, Pereira OL, Phillips AJL, Raspé O, Rollins AW, Romero AI, Etayo J, Selçuk F, Stephenson SL, Suetrong S, Taylor JE, Tsui CKM, Vizzini A, Abdel-Wahab MA, Wen TC, Boonmee S, Dai DQ, Daranagama DA, Dissanayake AJ, Ekanayaka AH, Fryar SC, Hongsanan S, Jayawardena RS, Li WJ, Perera RH, Phookamsak R, de Silva NI, Thambugala KM, Tian Q, Wijayawardene NN, Zhao RL, Zhao Q, Kang JC, Promputtha I (2015) The Faces of Fungi database: fungal names linked with morphology, phylogeny and human impacts. Fungal Diversity 74: 3–18. https://doi.org/10.1007/s13225-015-0351-8

Jeewon R, Hyde KD (2016) Establishing species boundaries and new taxa among fungi: recommendations to resolve taxonomic ambiguities. Mycosphere 7: 1669–1677. https://doi.org/10.5943/mycosphere/7/11/4

Jeewon R, Liew ECY, Hyde KD (2002) Phylogenetic relationships of *Pestalotiopsis* and allied genera inferred from ribosomal DNA sequences and morphological characters. Molecular Phylogenetics and Evolution 25: 378–392. https://doi.org/10.1016/S1055-7903(02)00422-0

Karunarathna A, Papizadeh M, Senanayake IC, Jeewon R, Phookamsak R, Goonasekara ID, Wanasinghe DN, Wijayawardene NN, Amoozegar MA, Shahzadeh Fazeli SA, Camporesi E, Hyde KD, Weerahewa HLD, Lumyong S, McKenzie EHC (2017) Novel fungal species of Phaeosphaeriaceae with an asexual/sexual morph connection. Mycosphere 8(10): 1818–1834. https://doi.org/10.5943/mycosphere/8/10/8

Katoh K, Rozewicki J, Yamada KD (2017) MAFFT online service: multiple sequence alignment, interactive sequence choice and visualization. Briefings in Bioinformatics 108: 1–7. https://doi.org/10.1093/bib/bbx108

Leuchtmann A (1984) Uber *Phaeosphaeria* Miyake und andere bitunicate Ascomyceten mit mehrfach querseptierten Ascosporen. Sydowia 37: 75–194. https://doi.org/10.3929/ethz-a-000320965

Maharachchikumbura SSN, Guo LD, Cai L, Chukeatirote E, Wu WP, Sun X, Crous PW, Bhat DJ, McKenzie EHC, Bahkali AH, Hyde KD (2012) A multi-locus backbone tree for *Pestalotiopsis*, with a polyphasic characterization of 14 new species. Fungal Diversity 56: 95–129. https://doi.org/10.1007/s13225-012-0198-1

Nylander JAA (2004) MrModeltest 2.0. Program distributed by the author. Evolutionary Biology Centre, Uppsala University.

Rambaut A, Drummond A (2008) FigureTree: Tree Figures drawing tool, version 1.2. 2. Institute of Evolutionary Biology, University of Edinburgh.

Ronquist F, Huelsenbeck JP (2003) MrBayes 3: Bayesian phylogenetic inference under mixed models. Bioinformatics 19: 1572–1574. https://doi.org/10.1093/bioinformatics/btg180

Shoemaker A, Babcock E (1989) *Phaeosphaeria*. Canadian Journal of Botany 67: 1500–1599. https://doi.org/10.1139/b89-199

Silvestro D, Michalak I (2012) raxmlGUI: a graphical front-end for RAxML. Organismic Diversity and Evolution 12: 335–337. https://doi.org/10.1007/s13127-011-0056-0

Stamatakis A, Alachiotis N (2010) Time and memory efficient likelihood-based tree searches on phylogenomic alignments with missing data. Bioinformatics 26: 1132–1139. https://doi.org/10.1093/bioinformatics/btq205

Tennakoon DS, Hyde KD, Phookamsak R, Wanasinghe DN, Camporesi E, Promputtha I (2016) Taxonomy and phylogeny of *Juncaceicola* gen. nov. (Phaeosphaeriaceae, Pleosporinae, Pleosporales). Cryptogamie, Mycologie 37(2): 135–156. https://doi.org/10.7872/crym/v37.iss2.2016.135

Thambugala KM, Camporesi E, Ariyawansa HA, Phookamsak R, Liu Z, Hyde KD (2014) Phylogeny and morphology of *Phaeosphaeriopsis triseptata* sp. nov., and *Phaeosphaeriopsis glaucopunctata*. Phytotaxa 176(1): 238–250. http://dx.doi.org/10.11646/phytotaxa.176.1.23

Wanasinghe DN, Phukhamsakda C, Hyde KD, Jeewon R, Lee HB, Jones EBG, Tibpromma S, Tennakoon DS, Dissanayake AJ, Jayasiri SC, Gafforov Y, Camporesi E, Bulgakov TS, Ekanayake AH, Perera RH, Samarakoon MC, Goonasekara ID, Mapook A, Li WJ, Senanayake IC, Li JF, Norphanphoun C, Doilom M, Bahkali AH, Xu JC, Mortimer PE, Tibell L, Savic ST, Karunarathna SC (2018) Fungal diversity notes 709–839: taxonomic and phylogenetic contributions to fungal taxa with an emphasis on fungi on Rosaceae. Fungal Diversity 89: 1–236. https://doi.org/10.1007/s13225-018-0395-7